Hydrolytic degradation of blends of poly(3-hydroxybutyrate) with poly(3-hydroxybutyrate-*co*-3-hydroxyvalerate)

Hidenori Satoh, Naoko Yoshie and Yoshio Inoue*
Department of Biomolecular Engineering, Tokyo Institute of Technology, 4259 Nagatsuta, Midori-ku, Yokohama 227, Japan

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The hydrolytic degradation behaviour and some other properties were analysed for blends of microbial poly(3-hydroxybutyrate) [P(3HB)] with microbial poly(3-hydroxybutyrate-*co*-3-hydroxyvalerate) [P(3HB-*co*-3HV); 3HV content, 22.3 mol%]. From the analysis of differential scanning calorimetry (d.s.c.) curves, the main component of the crystalline phase in the blend films was found to be P(3HB). The crystallinity of P(3HB) decreases on blending with P(3HB-*co*-3HV). The hydrolytic degradation of the blend films was carried out on the solution-cast films in phosphate buffer at 55°C and pH 7.4. The rate of hydrolytic degradation of the blend films was accelerated by blending with P(3HB-*co*-3HV), and showed linear correlation with crystallinity. Therefore, the degradation rate of P(3HB) can be controlled by blending with P(3HB-*co*-3HV). The lower the initial crystallinity of the blend films, the higher the rate of hydrolytic degradation is seen to be.

(Keywords: hydrolytic degradation; poly(3-hydroxybutyrate); blends)

INTRODUCTION

Poly(3-hydroxybutyrate) [P(3HB)] is a naturally occurring, crystalline, optically active polyester[1]. It is produced by bacterial fermentation and can be processed to yield several useful materials. P(3HB) is a thermoplastic degradable in the environment, by either hydrolytic or enzymatic degradation processes[2]; it is, however, reported to be relatively hard and brittle[3], and its melting behaviour is unstable with a narrow thermal processing window.

In order to obtain useful new derivatives based on P(3HB), two main approaches have been extensively studied. One is biosynthesis of copolyesters containing 3-hydroxyalkanoate units other than the 3HB unit with a variety of bacteria. Thus, for example, various 3HB copolymers containing several different hydroxyalkanoate units such as 3-hydroxyvalerate (3HV)[3], 4-hydroxybutyrate (4HB)[4], 5-hydroxybutyrate (5HB)[5], and 3-hydroxypropionate (3HP)[6] were bacterially synthesized. By comparison of the properties of these derivatives it has been shown that the P(3HB) copolymers with higher comonomer content are thermally more stable and exhibit progressively larger processing windows, and that the mechanical properties of P(3HB) can also be improved by the copolymerization.

The other method is by making miscible blends of P(3HB) and flexible biodegradable polymers or compounds. Avella and Martuscelli[7] reported the crystallization kinetics and morphology of a P(3HB)-based blend with poly(ethylene oxide). Holland *et al.*[8] have investigated the blends of P(3HB-*co*-3HV) and polysaccharides, and found that the hydrolytic degradation of P(3HB-*co*-3HV) is accelerated in the blends. It was therefore suggested that incorporation of soluble or hydrolytically unstable polymers such as polysaccharides results in substantial changes in the surface and bulk properties of the polymer matrix. In principle, blending of polymers may provide a wider range of morphologies, and hence of various physical properties, than does the use of each polymer system as a sole material. Many problems associated with the use of P(3HB) might be alleviated by use of blends of P(3HB) with P(3HB-*co*-3HV). Blends of P(3HB) with P(3HB-*co*-3HV) can produce either homogeneous or phase-separated morphologies[9].

The degradability of polymers has been evaluated by various methods. Doi *et al.*[10] investigated the hydrolytic and enzymatic degradation of P(3HB-*co*-3HV) and P(3HB-*co*-4HB). Besides thermal degradation[11], biodegradations in soil[12] and the sea[13] were also investigated. Fundamental study on the degradability of these polymers by hydrolysis is also thought to be important. Holland *et al.*[14] have investigated the hydrolytic degradation of P(3HB) and P(3HB-*co*-3HV) films prepared by various fabrication methods (solvent casting, hot pressing, injection moulding and cold compressing) at various pH values and temperatures. The rate of degradation was enhanced at higher temperature and accelerated at higher pH. Furthermore, molecular weight, copolymer structure and crystallinity affect the rate of hydrolytic degradation[8,15]. A change in crystallinity produces a difference in hydrolytic stability[16]. In the hydrolytic degradation of P(3HB-*co*-3HV), polymer surface changes may be detected[17] at an early stage. The hydrolysis occurs by a different pathway,

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* To whom correspondence should be addressed

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depending on whether the surrounding medium is alkaline or acidic[17]. The effect of crystallinity on the hydrolytic behaviour of polymer blend systems has been studied less. Hence, in this paper, we will report some properties of the blends of P(3HB) with P(3HB-*co*-3HV) and their hydrolytic degradation, and discuss the relation between degradability and crystallinity.

EXPERIMENTAL

Materials

P(3HB) and P(3HB-*co*-3HV) containing 22.3 mol% 3HV were used in this study. Both polymers were purchased from Aldrich Chemical Co. [P(3HB): lot no. 01703BV, $M_n = 77\,800$, $M_w/M_n = 2.17$; P(3HB-*co*-3HV): lot no. 06320PV, $M_n = 208\,000$, $M_w/M_n = 2.08$]. The 3HV content of the copolymer was determined by ^{1}H n.m.r. spectroscopy in chloroform. The P(3HB-*co*-3HV) sample has a random sequence distribution and is not a mixture of random copolymers with different comonomer compositions, which was confirmed by analysis of the ^{13}C n.m.r. spectrum observed in a chloroform solution[18].

Preparation of blend films

The films of P(3HB)/P(3HB-*co*-3HV) blends were prepared by casting from chloroform solutions. The resulting films were dried under vacuum at 50°C for at least 48 h until they reached constant weight. These films were aged for 3 weeks to reach equilibrium crystallinity prior to differential scanning calorimetry (d.s.c.) analysis and hydrolysis experiments. The compositions of the blends investigated here are summarized in *Table 1*.

Differential scanning calorimetry

The d.s.c. data were recorded on a Seiko DSC-20 equipped with an SSC-580 thermal controller. The films obtained by solution casting were heated from −50°C to 200°C at a heating rate of 10°C min^{-1}. The melting temperature (T_m) and heat of fusion (ΔH_m) were determined from the endothermic peak.

Hydrolytic degradation

Hydrolytic degradation of the blend films was carried out in phosphate buffer at 55°C and pH 7.4. Each film (20 mm × 15 mm, 5–10 mg) was placed in a test tube containing 15 ml phosphate buffer and shaken at 55°C. Wet gravimetric weight (W_w) of the film was determined periodically after removing the sample from the test tube, washing it in distilled water and surface drying with filter paper. The samples were dried *in vacuo* for 48 h to constant weight and reweighed to obtain the dry weight (W_d).

Gel permeation chromatography (g.p.c.)

Molecular weight was determined by g.p.c. performed at 40°C on a Toso HLC-8020 GPC system with a Toso SC-8010 controller and a refractive detector with TSK gel G2000H$_{XL}$ and GMH$_{XL}$ columns. Chloroform was used as the eluant at a flow rate of 1.0 cm^3 min^{-1}, and sample concentration was 0.5 mg cm^{-3}. Polystyrene standards of low polydispersity were used to construct a calibration curve. G.p.c. data were processed on a SC-8010 data processer to calculate average molecular weights, M_n and M_w, as well as polydispersity, M_w/M_n.

Table 1 Weight composition of P(3HB)/P(3HB-*co*-3HV) blends

Sample code	P(3HB)/P(3HB-*co*-3HV)
1	100/0
2	75/25
3	50/50
4	25/75

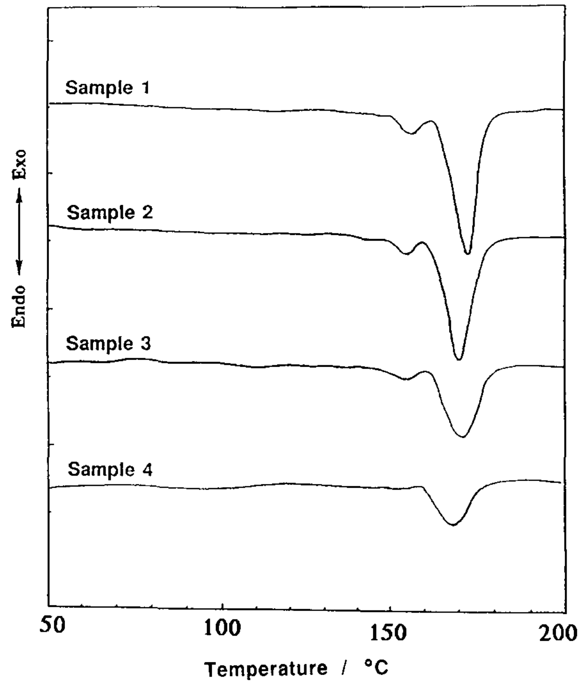

Figure 1 D.s.c. thermograms of blend films

RESULTS AND DISCUSSION

Some properties of the blend films were found to change gradually with changing P(3HB-*co*-3HV) content. In appearance, P(3HB) film (sample 1) was opaque and brittle, while the blend films (samples 2–4) became clearer and softer with increasing P(3HB-*co*-3HV) content.

Thermal properties

The d.s.c. thermograms of the blend films obtained by solution casting are shown in *Figure 1*. Each blend film shows one main endothermic peak when heated from −50°C to 200°C. As each thermogram showed one main peak, the blend was thought to consist of a single crystalline phase. Furthermore, the melting temperatures of samples 2–4 were close to that of P(3HB) homopolymer. Consequently, the main component of the crystalline phase is P(3HB). The melting temperature and heat of fusion of the P(3HB) phase in the blend samples are given from this peak. *Figure 2* shows the plot of the melting temperature against the P(3HB-*co*-3HV) content. It is known that the melting temperature of the P(3HB) phase continuously decreases as P(3HB-*co*-3HV) content increases. *Figure 3* shows the plot of the heat of fusion against the P(3HB-*co*-3HV) content. The heat of fusion of the blends decreases reciprocally to the increase in P(3HB-*co*-3HV) content. Because P(3HB-*co*-3HV) cannot form a crystalline phase, the decrease of heat of fusion with increasing P(3HB-*co*-3HV) content could be well understood. The heat of fusion relative to the unit P(3HB) content seems to be almost constant. The crystallinity of the P(3HB) phase is calculated from the

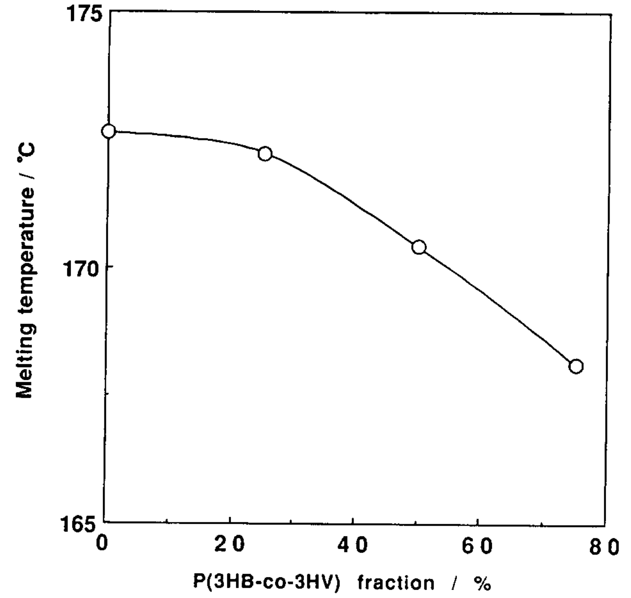

Figure 2 Plot of melting temperature *versus* fraction of P(3HB-*co*-3HV) (%) for blend films

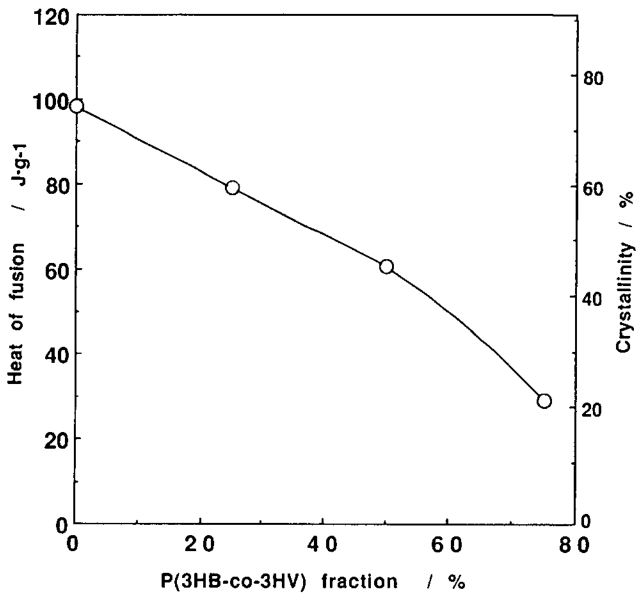

Figure 3 Plot of heat of fusion and crystallinity *versus* fraction of P(3HB-*co*-3HV) for blend films

heat of fusion observed here, by assuming the heat of fusion for 100% P(3HB) crystal to be 11.2 kJ mol^{-1} (ref. 19). While the crystallinities of the single polymer systems of P(3HB) and P(3HB-*co*-3HV) are as high as 72.5%and 39.5%, respectively, the crystallinity of the blend films decreases with increasing P(3HB-*co*-3HV) content. Thus, this blend system has a lower crystallinity than P(3HB). The crystallinity of sample 4 is 22.4%. These data are important in that the rate of hydrolytic degradation is expected to be affected by the crystallinity of the blend films.

Hydrolytic degradation

The changes of wet and dry weights were calculated by using the following equations:

$$\text{Wet weight change} = [W_{w(t)} - W_{d(0)}]/W_{d(0)}$$

$$\text{Dry weight change} = [W_{d(t)} - W_{d(0)}]/W_{d(0)}$$

where $W_{w(t)}$, $W_{d(t)}$, and $W_{d(0)}$ are wet weight at time t, dry weight at time t, and initial dry weight, respectively. *Figure 4* plots the changes of wet and dry weight against the degradation time for sample 2. The plots for the other samples indicated the same tendency. The changes of wet and dry gravimetric weight show a considerable difference. While the wet weight initially increased slightly, mainly due to hydration, and subsequently became constant, the dry weight was unchanged for a while. The disintegration and weight loss of sample films occurred after 80, 100, 120 and 150 days hydrolysis for samples 1, 2, 3 and 4, respectively. At this time, the number-average molecular weight of each sample film is less than 5.00×10^4. With increasing P(3HB) content, the sample films were broken at an earlier stage.

Molecular weight change

From the g.p.c. curves, the number-average molecular weight (M_n) and polydispersity (M_w/M_n) were calculated. The value of M_w/M_n was kept approximately constant during hydrolytic degradation. *Figure 5* shows the plots of M_n against degradation time for sample films with different blend ratios. The values of M_n were reasonably reproducible and provided a good measure for understanding the degradation process. All of the samples showed a continous drop in M_n with degradation time, while the values of M_w/M_n were approximately constant for each sample. The average number, N, of bond cleavages per single polymer chain at time t, is given by:

$$N = M_{n(0)}/M_{n(t)} - 1$$

where $M_{n(0)}$ and $M_{n(t)}$ are number-average molecular weight at degradation times 0 and t, respectively. *Figure 6* shows a relationship between the N value and time t for samples 1–4. It is suggested that the number of bond cleavages gradually increases with progress of hydrolytic degradation. The rate of hydrolytic degradation is apparently influenced by the blend ratio, and it decreases with increasing P(3HB) content, in the sequence sample 4 > sample 3 > sample 2 > sample 1.

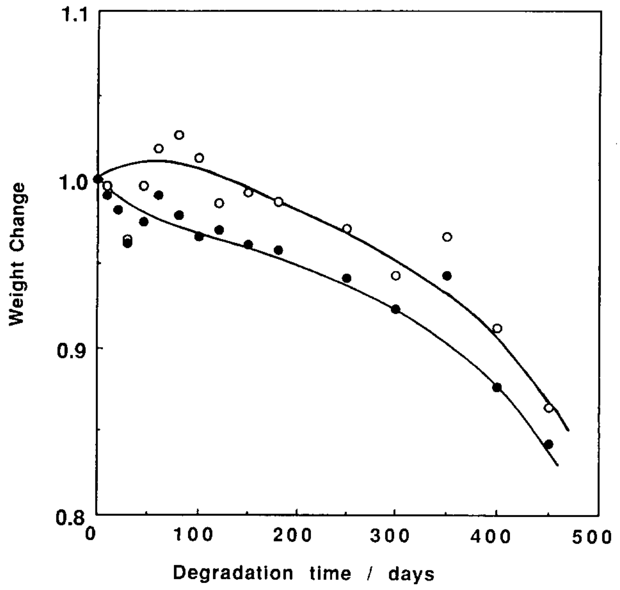

Figure 4 Wet (○) and dry (●) weight changes of sample 2 during hydrolytic degradation in phosphate buffer at 55°C and pH 7.4

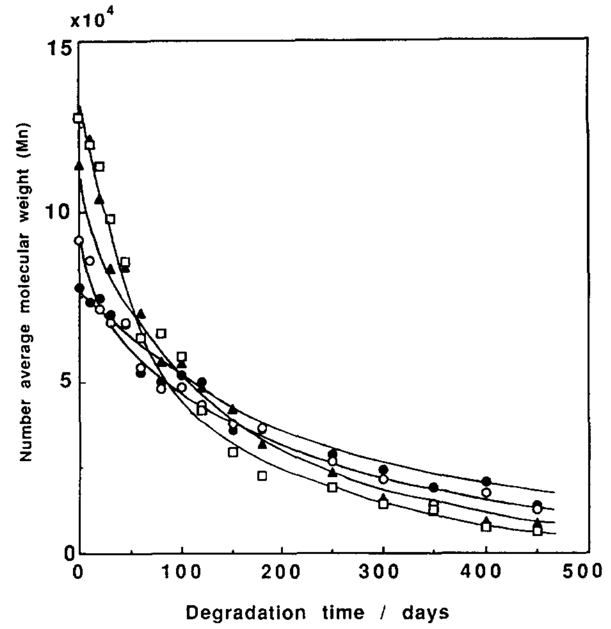

Figure 5 Changes in the M_n values of blend films during hydrolytic degradation in phosphate buffer at 55°C and pH 7.4, for sample 1 (●), sample 2 (○), sample 3 (▲) and sample 4 (□)

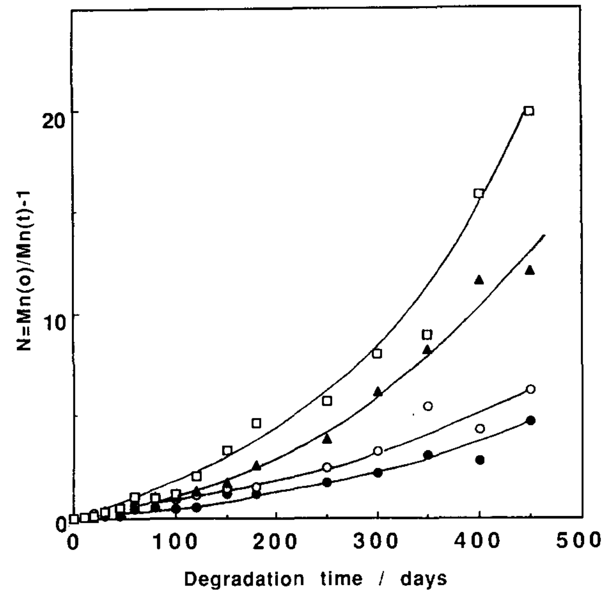

Figure 6 Plots of $[M_{n(0)}/M_{n(t)}]-1$ *versus* degradation time t for blend films: sample 1 (●); sample 2 (○); sample 3 (▲); sample 4 (□)

The changes in the value of N are not linear but almost fit an exponential function curve. *Figure 7* shows plots of $\log[M_{n(t)}/M_{n(0)}]$ against degradation time t. If the chain scission occurs at random, the value of $\log\{M_{n(t)}/M_{n(0)}\}$ would be a linear function of time t. As anticipated, *Figure 7* shows a linear relationship, indicating random chain scission. The rate constant of the first-order hydrolytic degradation, k, is given by

$$k=-d[\log\{M_{n(t)}/M_{n(0)}\}]/dt$$

The calculated degradation rate constants k are 1.648, 1.992, 2.691 and 3.065 for samples 1, 2, 3 and 4, respectively. Plots of the value k against crystallinity are illustrated in *Figure 8*, indicating a linear relationship between the values: the lower the initial crystallinity in the blend films, the higher rate of hydrolytic degradation. Since the crystallinity decreases with increase of P(3HB-*co*-3HV) content, the crystallinity and degradation rate of P(3HB) blends can be controlled by adjusting the P(3HB-*co*-3HV) content.

Changes in thermal properties

Changes in melting temperature, heat of fusion and crystallinity were determined using d.s.c. In *Figure 9* are shown the plots of melting temperature against the degradation time t, indicating continuous decrease in melting temperature for each sample with the progress of degradation. These changes of melting temperature seem to result from decrease in molecular weight. *Figure 10* shows the plots of heat of fusion and crystallinity against degradation time t. The crystallinity decreased initially for the first 60 days and then became

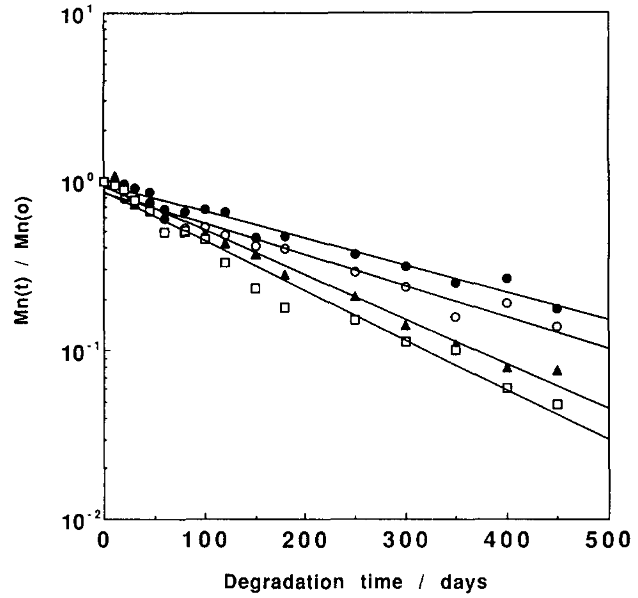

Figure 7 Changes in the $\log\{M_{n(t)}/M_{n(0)}\}$ values of blend films during hydrolytic degradation in phosphate buffer at 55°C and pH 7.4: sample 1 (●); sample 2 (○); sample 3 (▲); sample 4 (□)

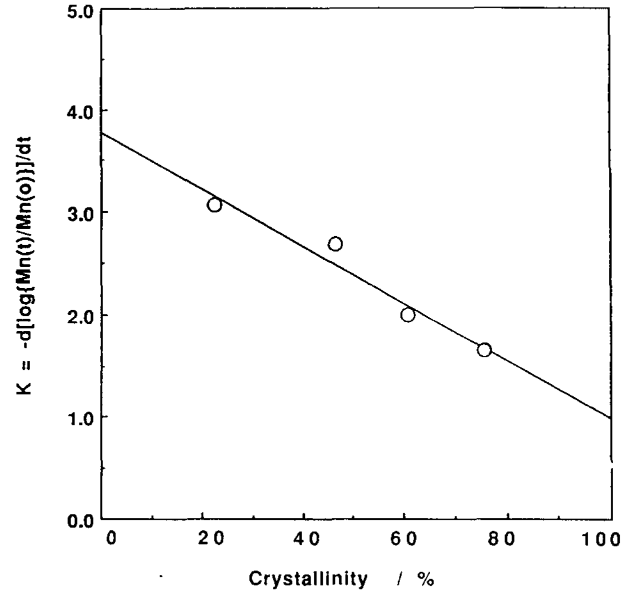

Figure 8 Plot of $-d[\log\{M_{n(t)}/M_{n(0)}]/dt$ *versus* crystallinity for blend films during hydrolytic degradation in phosphate buffer at 55°C and pH 7.4

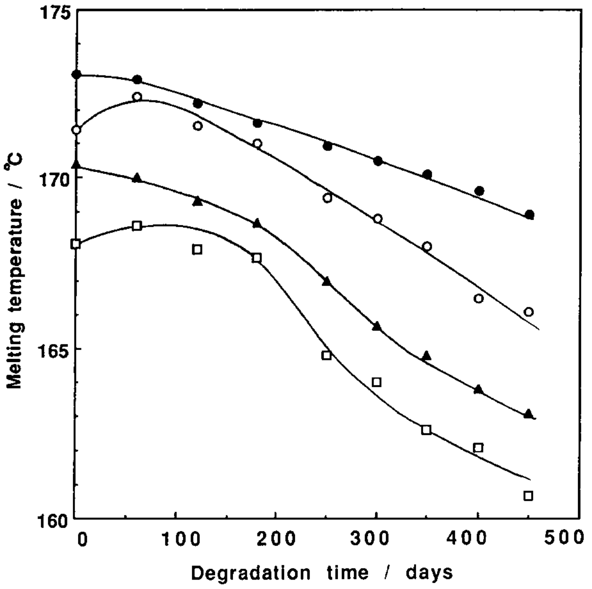

Figure 9 Plots of melting temperature *versus* degradation time *t* for blend films during hydrolytic degradation in phosphate buffer at 55°C and pH 7.4: sample 1 (●); sample 2 (○); sample 3 (▲); sample 4 (□)

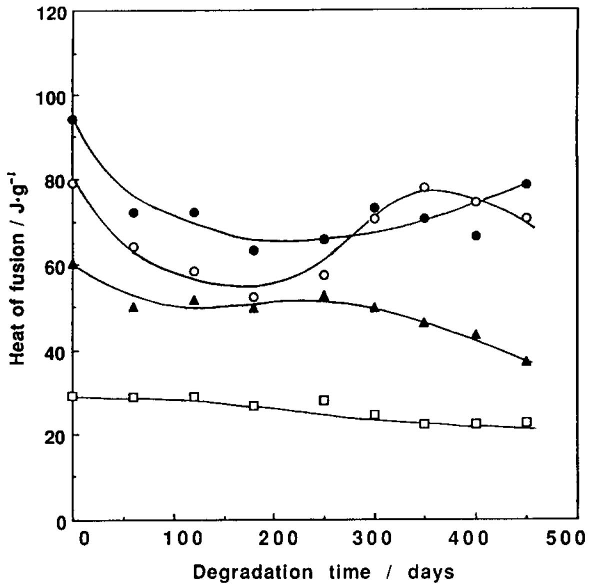

Figure 10 Plots of heat of fusion *versus* degradation time *t* for blend films during hydrolytic degradation in phosphate buffer at 55°C and pH 7.4: sample 1 (●); sample 2 (○); sample 3 (▲); sample 4 (□)

constant for a while. The decreases in crystallinity for the first 60 days were 17.5%, 11.3%, 7.8% and 0.3% for samples 1, 2, 3 and 4, respectively. However, the crystallinities, in particular that of sample 2, apparently increased at about 250 days. This increase in crystallinity correlated well with the profiles of the gravimetric weight loss. As a rule, the crystallinity decreases initially and increases secondarily with hydrolytic degradation. The first-stage decrease of crystallinity results from the disintegration of the crystalline phase in aqueous solution, and the latter-stage increase results from erosion of the amorphous region of the blend films.

CONCLUSIONS

The crystallinity of blends of P(3HB) with P(3HB-*co*-3HV) can be controlled by adjusting P(3HB-*co*-3HV) content. The degradation rate has a linear relationship with the crystallinity. Hence, the degradation rate of films of this blend can be controlled, because the amorphous phase is degraded more rapidly than the crystalline phase. Although properties other than hydrolytic degradability remain largely unexplored, such blends probably offer the best potential to produce commercially viable materials.

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